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SOME AEROPHYTIC ALGAE IN THE „MOKRY BÓR” RESERVE

GLONY AEROFITYCZNE W REZERWACIE „MOKRY BÓR”

ABSTRACT: the results of investigation on aerophytic algae grown on different tree species in the area of the „Mokry Bór” reserve carried in February 1995. Among the found green algae *Apatococcus lobatus* played important role. It occurred on the surface of nearly all examined trees creating dark green coatings on the tree barks (excluding *Carpinus betulus*). On *Abies* and *Picea* apart from *Apatococcus* also other synantropic green alga occurred *Gloeotila protogenita*. They were accompanied by the following species: *Chlorella vulgaris*, *Ch. ellipsoidea*, *Klebsormidium flaccidum*, *Stichococcus bacillaris*, *Pseudococcomyxa simplex* and *Heterothrix exilis*, representing probably previous natural communities.

Obtained ultrastructures of cells of *Apatococcus* showed inside some specimens presence of fungus haustorium, which however did not affect cellular structures of this algae. This form of symbiosis has not been observed so far in the case of this green alga. Characteristic ultrastructure observed in the green alga *Gloeotila* (in particular the thick layer of mucus surrounding the vegetative cells) proves presence on this area of a typical land form containing pyrenoid of a specific structure. Its land character is also proven by the reproduction ways.

KEY WORDS: aerophytic algae, synantropisation, haustorium, ultrastructures.

Contents

1. Introduction
2. Materials and methods
3. Results and conclusion
4. Discussion
5. Literature cited
5. Streszczenie

1. INTRODUCTION

Aerophytic flora of algae on the area of the Świętokrzyski National Park is still not known to great extent. The changes are observed in the structure of forest stands and the setting of natural ecosystems. A good example here may be the area of the "Mokry Bór" reserve, which is deprived on the large area of lagging which makes it exposed to direct contact with the motorway and village buildings. High level of human interference may be proven by high level of synantropisation, massive regression of mosses and lichens that are substituted by algae. In such a situation the results of investigation of epiphytic microflora of trees, are of special importance.

2. MATERIALS AND METHODS

Aerophytic algae were collected in February 1995 at strict reserve "Mokry Bór", which is one of five reserves in the area of the Świętokrzyski National Park (Fig. 1). It is located in Wilkowska valley and covers the area of 37.9 ha.

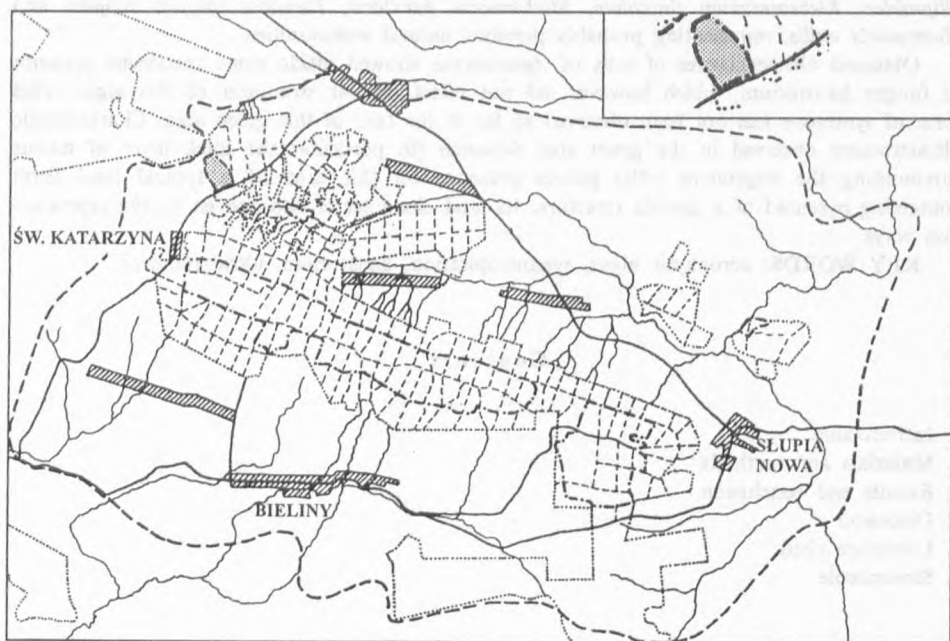


Fig. 1. Map of "Mokry Bór" reserve within Świętokrzyski National Park
Rys. 1. Rezerwat „Mokry Bór” (Świętokrzyski Park Narodowy)

Material was removed from the surface of tree bark and cultured on the standard Bristol agar medium, at 20 C under a 12/12 h light/dark cycle at 3000 lx provided by 40W fluorescent tubes.

For transmission electron microscopy the laboratory-grown cells were removed with a spatula from the surface of agar plates, and dispersed in fixative. The initial fixative was 2.6% glutaraldehyde and 0.05% ruthenium red in 0.067 M phosphate buffer at pH 7.1 applied at room temperature for 1 h. Three washes of 5 min each were done in buffer. The second fixative, applied also at room temperature, was 1.33% osmium tetroxide and 0.05% ruthenium red in buffer. Mixing of the osmium tetroxide and ruthenium red was done immediately prior to use. Such treatment immobilizes and stains a maximum amount of secreted mucilage enclosing the cells (Luft 1971). After several subsequent washes in buffer and dehydration in ethanol series, the cells were infiltrated over-night with a 1 : 1 mixture of propylene oxide and SPURR's hard medium (1969), and then embedded in the fresh SPURR's medium. The samples were polymerized at 60 C for 18 h. The sections were cut with glass knives on a Reichert-Jung ultramicrotome. Observations and photographs were made with a Tesla BS 500 electron microscope.

3. RESULTS AND CONCLUSION

During the investigation performed in February 1995 at strict reserve „Mokry Bór”, on different trees growing on that area 8 species of aerophytic algae from the groups such as *Chlorophyceae*, *Charophyceae* and *Xanthophyceae* were found (Tab. I). Among them important role was played by *Apatococcus lobatus* (Figs. 2-3). It was found on the surface of majority of investigated trees (*Pinus*, *Picea*, *Abies*, *Quercus*, *Populus* and *Betula*). It was not found only on the *Carpinus betulus*.

It densely covered especially barks of *Pinus* trees growing closer to the motorway, creating thick, dark green coatings. On the specimens growing inside the reserve, at some distance from the road, thallus of *Apatococcus* did not occur in the form of compact coating, and also some amount of *Chlorella ellipsoidea* were observed, which is probably remnant of natural community. On other trees, mentioned above, the number of species occurring together with *Apatococcus* and representing primary communities was significantly greater (Tab. I).

On *Picea* and *Abies* the effect obtained by the former green algae was additionally reinforced thanks to specimens of *Gloeotila protogenita* (Figs. 6-9). It is interesting that both mentioned species are characteristic for environments affected by human operations. *Apatococcus* is common also on urbanized areas while *Gloeotila* occurs in masses on roofs of village buildings.

Table I

List of aerophytic algae found on different tree species in „Mokry Bór” reserve (Świętokrzyski National Park)
 Lista gatunków glonów aerofitycznych znalezionych na korze różnych gatunków drzew w rezerwacie ścisłym „Mokry Bór”
 (Świętokrzyski Park Narodowy)

Taxa	<i>Pinus silvestris</i> L.	<i>Picea excelsa</i> (Lam.) L.	<i>Abies alba</i> Mill.	<i>Quercus sessilis</i> Ehr.	<i>Carpinus betulus</i> L.	<i>Betula verrucosa</i> Ehr.	<i>Populus tremula</i> L.
<i>Chlorophyceae</i>							
<i>Apatococcus lobatus</i> (Chodat) Boye-Pet.	+	+	+	+		+	+
<i>Chlorella ellipsoidea</i> Gem.	+	+		+			
<i>Ch. vulgaris</i> Beij		+		+			
<i>Gloeotila protogenita</i> Kuetz.		+	+				
<i>Pseudococcomyxa simplex</i> (Mar.) Fott		+	+	+			
<i>Charophyceae</i>							
<i>Klebsormidium flaccidum</i> (Kuetz.) Silva			+		+	+	+
<i>Stichococcus bacillaris</i> Naeg.		+	+	+		+	+
<i>Xanthophyceae</i>							
<i>Heterothrix exilis</i> (Klebs) Pascher				+	+	+	

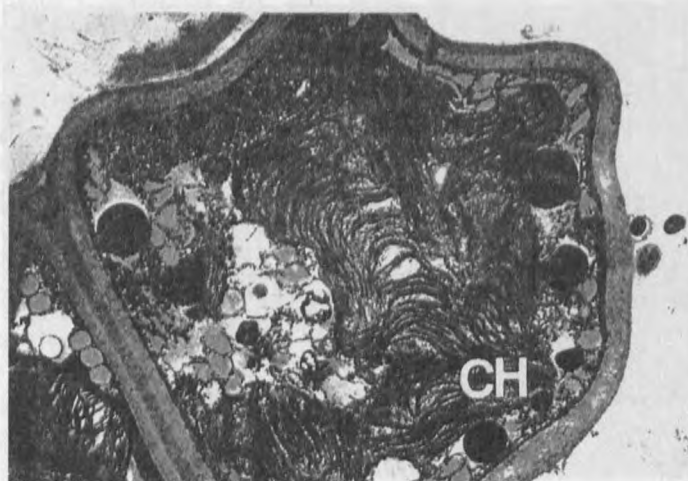


Fig. 2. *Apatococcus lobatus* (Chodat) Boye-Pet. (transmission electron microscope). Part of vegetative cell; Ch, chloroplast; x15 000

Rys. 2. *Apatococcus lobatus* (Chodat) Boye-Pet. (transmisyjny mikroskop elektronowy). Fragment komórki wegetatywnej; Ch, chloroplast; x15 000

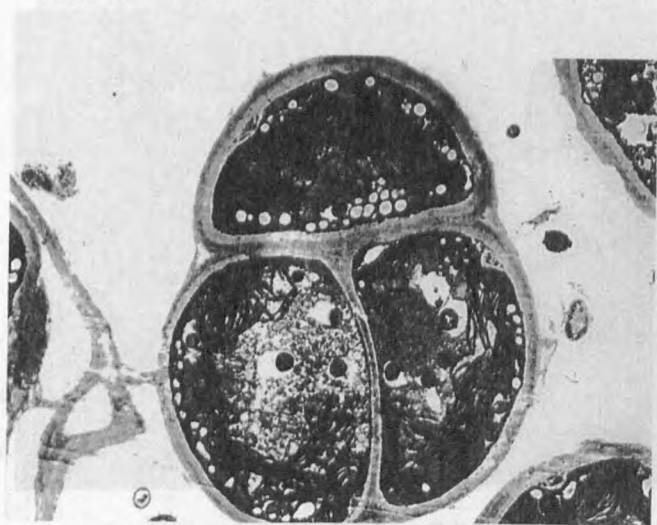


Fig. 3. *Apatococcus lobatus* (Chodat) Boye-Pet. (transmission electron microscope). Cell aggregation x9000

Rys. 3. *Apatococcus lobatus* (Chodat) Boye-Pet. (transmisyjny mikroskop elektronowy). Skupienie komórek; x9000

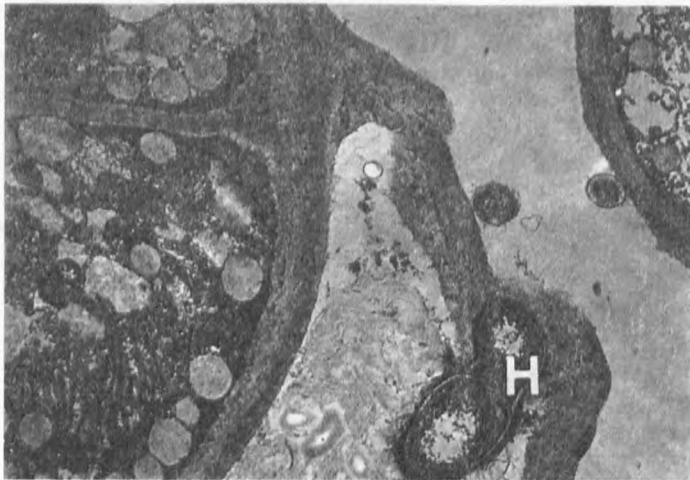


Fig. 4. *Apatococcus lobatus* (Chodat) Boye-Pet. (transmission electron microscope). Part of vegetative cell with haustorium; H, haustorium; x11 000

Rys. 4. *Apatococcus lobatus* (Chodat) Boye-Pet. (transmisyjny mikroskop elektronowy). Fragment komórki wegetatywnej z haustorium; H, haustorium; x11 000

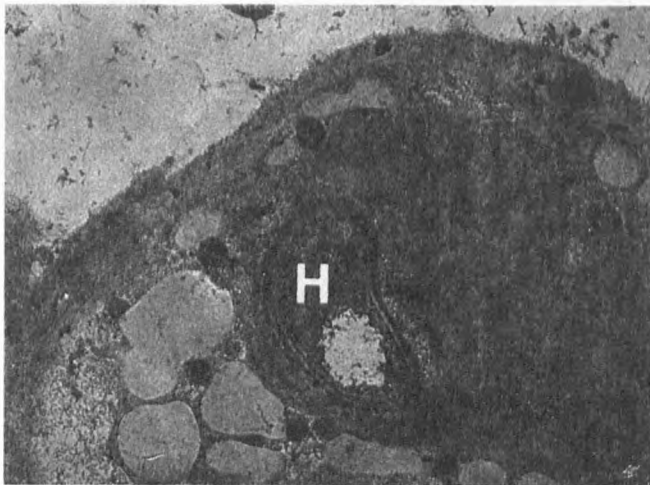


Fig. 5. *Apatococcus lobatus* (Chodat) Boye-Pet. (transmission electron microscope). Haustorium inside the enlarged vegetative cell; x16 000

Rys. 5. *Apatococcus lobatus* (Chodat) Boye-Pet. (transmisyjny mikroskop elektronowy). Haustorium wewnątrz powiększonej komórki wegetatywnej; x16 000

The changes observed on this area, especially within the epiphytic microflora of trees in the form of appearance of new floral elements and partial reduction of elements of primal flora, well illustrate the transformations taking place in this area as a result of human activities. Lack of natural lagging zone on the significant length of the reserve as well as direct contacts with the motorway and village building is probably the main reason of changes. Formation of new communities on the trees with *Apatococcus* and *Gloetila* as dominating species proves the direction and quality of changes. It is also interesting to observe linking of fungus with *Apatococcus*. Analysis of ultrastructures of cells of these algae proved in some of the specimens presence of haustorium of the fungus, which were not affecting cell structures of the algae (Figs. 4-5). It allows for the opinion that a new form of symbiosis occurred here between the algae which has not been observed so far in such a state and in the case of this fungus.



Fig. 6. *Gloetila protogenita* Kütz. (transmission electron microscope). 4 autospores inside the mother cell; x12 000

Rys. 6. *Gloetila protogenita* Kütz. (transmisyjny mikroskop elektronowy). 4 autospory wewnątrz komórki macierzystej; x12 000

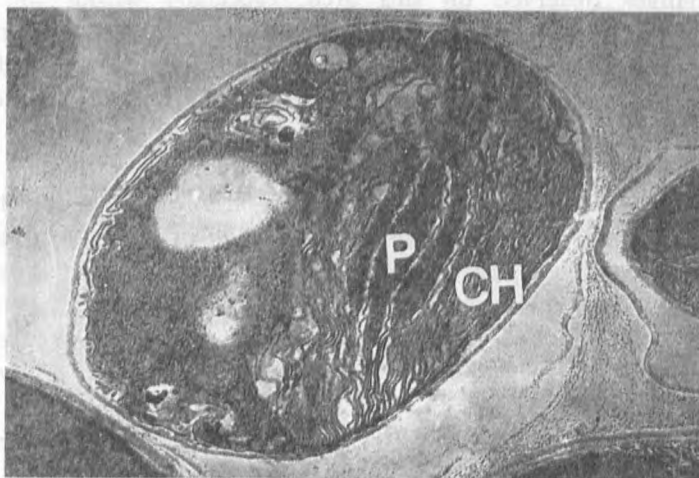


Fig. 7. *Gloetila protogenita* Kutz. (transmission electron microscope). Oblong vegetative cell; P, pyrenoid; Ch, chloroplast; $\times 15\,000$

Rys. 7. *Gloetila protogenita* Kutz. (transmisyjny mikroskop elektronowy). Podłużna komórka wegetatywna. P, pirenoid; Ch, chloroplast; $\times 15\,000$

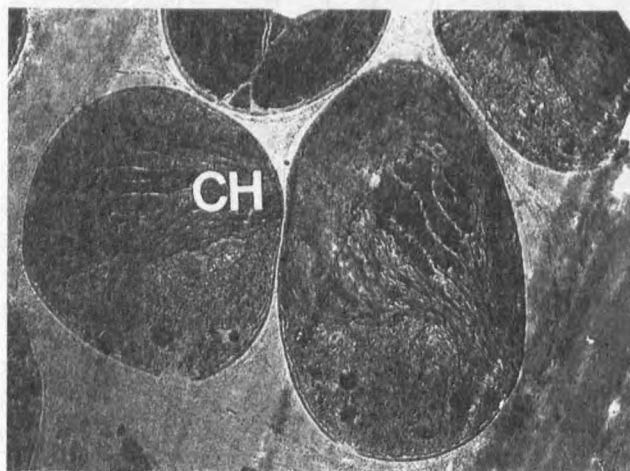


Fig. 8. *Gloetila protogenita* Kutz. (transmission electron microscope). Aplanospores; P, pyrenoid; Ch, chloroplast; $\times 11\,500$

Rys. 8. *Gloetila protogenita* Kutz. (transmisyjny mikroskop elektronowy). Aplanospory; P, pirenoid; Ch, chloroplast; $\times 11\,500$

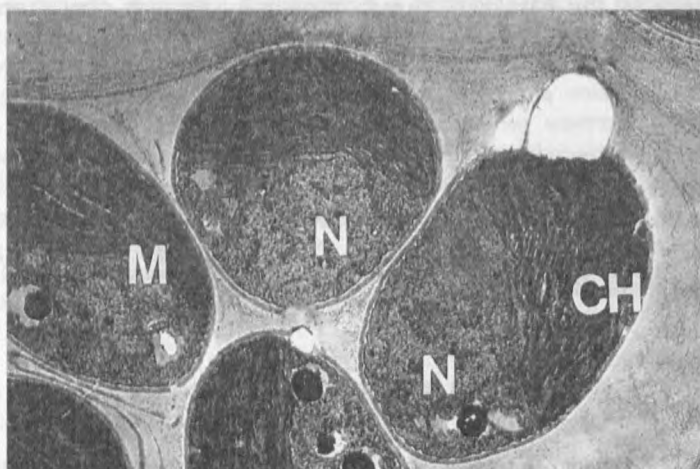


Fig. 9. *Gloetila protogenita* Kutz. (transmission electron microscope). Aplanospores; M, mitochondrium; N, P, nucleus; Ch, chloroplast; x12 500

Rys. 9. *Gloetila protogenita* Kutz. (transmisyjny mikroskop elektronowy). Aplanospory; M, mitochondrium; N, jądro; Ch, chloroplast; x12 500

Mentioned above green alga *Gloetila protogenita* (Figs. 6–9) is a typical terrestrial alga in this area. Its terrestrial character is proven also by the way of its reproduction (by means of aplanospores). It is also stressed by the presence of thick layer of mucus which is clearly visible on electrogrammes. This mucus probably protects the cells against drying.

4. DISCUSSION

Some progress may be observed in studies on aerophytic flora occurring on tree barks surface (Gartner 1994, Gartner & Ingolić 1989, Mrozińska 1990, 1991, 1992). Our knowledge on changes in this flora caused especially by human operations is also still limited.

Aerophytic flora on the area of „Mokry Bór” reserve is relatively scarce if compared with flora of Białowiecki National Park (Mrozińska 1992), however it is very interesting. The presence of two synantropic species, *Apatococcus labatus* and *Gloetila protogenita*, occurring frequently with small number of species representing natural environment, proves significant changes happened in this area. In some cells of *Apatococcus* haustoria a fungus has been found so far not spotted in the case of this alga which

also defines the range of changes. Reducation of aerophytic algae flora on this area goes in the part with direct impact by human operation, towards domination of *Apatococcus*.

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5. LITERATURE CITED

- Gartner G. 1994. Zur Taxonomie aerophiler grüner Algenanflüge an Baumrinden. Ber. Nat.-Med. Verien Innsbruck, 81: 51–59.
- Gartner G., Ingolić E. 1989. Ein Beitrag zur Kenntnis von *Apatococcus lobatus* (Chlorophyta, Chaetophorales). Pl. Syst. Evol., 164: 133–143.
- Luft J. H. 1971. Ruthenium red and violet. I. Chemistry, purification, methods of use for electron microscopy and mechanism of action. Anat. Rec., 171: 347–368.
- Mrozińska T. 1990. Aerophytic algae from North Korea. Algological Stud., 58: 29–47.
- Mrozińska T. 1991. Algological guide through Częstochowa Highlands on the background of past geological periods and presently. PAN, Kraków: 3–32.
- Mrozińska T. 1992. Aerophytic Algae. Phytocoenosis 4. Archivum Geobotanicum 3. Warszawa-Białowieża: 45–47.
- Spurr A. R. 1969. A low-viscosity epoxy resin embedding medium for electron microscopy. J. Ultrastruct. Res., 26: 31–43.

6. STRESZCZENIE

W tej pracy przedstawione zostały wyniki badań dotyczące glonów aerofitycznych, występujących na różnych drzewach na terenie rezerwatu ścisłego „Mokry Bór” (Świątokrzyski Park Narodowy). Materiał został zebrany w lutym 1995 r. Stwierdzono na prawie wszystkich badanych drzewach (z wyjątkiem *Carpinus betulus*) obecność *Apatococcus lobatus*.

Na powierzchni okazów *Picea*, rosnących na obrzeżu rezerwatu, tuż koło drogi, *Apatococcus* tworzył jednolite pokrywy. Jedynie na okazach *Picea* rosnących wewnątrz rezerwatu występowały poza nim skupienia *Chlorella ellipsoidea*. Natomiast na korze pozostałych drzew obserwowano znacznie większą liczbę gatunków towarzyszących, będących prawdopodobnie pozostałościami poprzednich, naturalnych zbiorowisk aerofitycznych. Są to: *Stichococcus bacillaris*, *Pseudococcomyxa simplex*, *Chlorella ellipsoidea*, *Ch. vulgaris*, *Klebsormidium flaccidum* i *Heterothrix exilis*. Poza tym na *Abies* i *Picea* występował jeszcze drugi mało znany gatunek synantropijny *Gloeotila protogenita* o charakterze glonu lądowego. Świadczy o tym gruba warstwa śluzu wokół komórek oraz jej sposób rozmnażania.

Obserwowane na tym terenie zmiany, zachodzące w obrębie mikroflory drzew w formie pojawienia się nowych elementów florystycznych i częściowej redukcji elementów pierwotnej flory, dobrze obrazują przekształcenia i kierunek zmian. Brak na znacznej długości rezerwatu naturalnej strefy otulinowej, jak również jego bezpośredni kontakt z drogą szybkiego ruchu i zabudowaniami wiejskimi są prawdopodobnie główną przyczyną zmian.

Analiza ultrastruktur komórek *Apatococcus* wykazała we wnętrzu niektórych okazów obecność haustorii grzyba, nie naruszającego struktur komórkowych tego glonu. Można wnioskować, że powstała tutaj nowa forma symbiozy między tym glonem a grzybem, dotychczas nie obserwowana u tego gatunku.

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